

Appn. No.09/890,297
Amdt. Dated April 18, 2003
Reply to Office action of February 20, 2003

Attorney Fil No.: CE0253 US

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

Claims 1-53 (cancelled)

Claim 54 (new): A process for purifying an albumin solution, the process comprising

- (1) subjecting the albumin solution to cation exchange chromatography in the negative mode with respect to the albumin in order to yield an albumin-containing cation exchange product;
- (2) subjecting the albumin-containing cation exchange product, with or without intervening purification steps, to anion exchange chromatography to yield an albumin-containing anion exchange product; and
- (3) placing the albumin-containing anion exchange product, without further purification, into a final container for therapeutic use

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Claim 55 (new): A process according to Claim 54 wherein the initial albumin solution contains glycosylated albumin and the glycosylated albumin is bound during the said cation exchange step.

Claim 56 (new): A process according to Claim 54 wherein the cation exchange step utilises a matrix which comprises immobilised sulfopropyl substituents as cation exchangers.

Claim 57 (new): A process according to Claim 54 wherein the initial albumin solution has a pH of 4.5-6.0.

Claim 58 (new): A process according to Claim 54 wherein the initial albumin solution has an albumin concentration of 10-250g.L⁻¹.

Claim 59 (new): A process according to Claim 54 wherein the initial albumin solution has an octanoate ion concentration of 2-15mM.

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Claim 60 (new): A process according to Claim 54 wherein the initial albumin solution has an octanoate ion concentration of 5-10mM.

Claim 61 (new): A process according to Claim 54 wherein prior to the cation exchange step the albumin solution undergoes at least one step selected from the group consisting of: (i) pH-adjustment; (ii) concentration; (iii) diafiltration; and (iv) conditioning by addition of a fatty acid.

Claim 62 (new): A process according to Claim 61 wherein prior to the cation exchange step the albumin solution undergoes conditioning by addition of an octanoate salt.

Claim 63 (new): A process according to Claim 54 wherein the anion exchange step utilises a matrix which comprises immobilised dialkylaminoalkyl substituents as anion exchangers.

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Claim 64 (new): A process according to Claim 54 wherein the anion exchange step is run in the negative mode with respect to the albumin.

Claim 65 (new): A process according to Claim 64 wherein the albumin solution which undergoes anion exchange chromatography has a pH of 4.0-5.2.

Claim 66 (new): A process according to Claim 64 wherein the albumin solution which undergoes anion exchange chromatography has a conductivity of less than 4.0mS.cm⁻¹.

Claim 67 (new): A process according to Claim 54 wherein the anion exchange step is run in positive mode with respect to the albumin.

Claim 68 (new): A process according to Claim 67 wherein the albumin solution which undergoes positive mode anion exchange chromatography has a pH of 6.0-8.0.

Claim 69 (new): A process according to Claim 67 wherein the concentration of the albumin in the albumin solution which undergoes positive mode anion exchange chromatography is 10-100g.L⁻¹

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~~Claim 70 (new): A process according to Claim 67 wherein the albumin solution which undergoes positive mode anion exchange chromatography has a conductivity of 1.0-1.5mS.cm⁻¹.~~

~~Claim 71 (new): A process according to Claim 67 wherein the albumin is eluted in the anion exchange step using a buffer comprising a compound having a specific affinity for albumin.~~

~~Claim 72 (new): A process according to Claim 71 wherein the buffer comprises 20-90mM phosphoric acid salt.~~

~~Claim 73 (new): A process according to Claim 67 wherein the albumin is eluted in the anion exchange step with a buffer of pH6.0-8.0.~~

~~Claim 74 (new): A process according to Claim 54 wherein, prior to the anion exchange step, the albumin solution undergoes at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning~~

~~Claim 75 (new): A process according to Claim 54 wherein the process is preceded by at least one step selected from the group consisting of: fermentation; primary separation; concentration; conditioning; cation exchange chromatography; anion exchange chromatography; and affinity chromatography.~~

~~Claim 76 (new): A process for purifying an albumin solution, the process comprising~~

- ~~(1) subjecting the albumin solution to cation exchange chromatography in the negative mode with respect to the albumin in order to yield an albumin-containing cation exchange product;~~
- ~~(2) subjecting the albumin-containing cation exchange product, with or without intervening purification steps, to anion exchange chromatography to yield an albumin-containing anion exchange product; and~~

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~~(3) placing the albumin-containing anion exchange product, without further purification, into a final container for therapeutic use,~~
wherein the albumin-containing anion exchange product is subjected to at least one step selected from the group consisting of buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning, before being placed into the said final container.

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Claim 77 (new): A process according to Claim 76 wherein the initial albumin solution contains glycosylated albumin and the glycosylated albumin is bound during the said cation exchange step.

Claim 78 (new): A process according to Claim 76 wherein prior to the cation exchange step the albumin solution undergoes at least one step selected from the group consisting of: (i) pH-adjustment; (ii) concentration; (iii) diafiltration; and (iv) conditioning by addition of a fatty acid.

Claim 79 (new): A process for purifying an albumin solution, the process comprising
(1) subjecting the albumin solution to anion exchange chromatography in order to yield an albumin-containing anion exchange product;
(2) subjecting the albumin-containing anion exchange product, with or without intervening purification steps, to cation exchange chromatography run in the negative mode with respect to the albumin to yield an albumin-containing cation exchange product; and
(3) placing the albumin-containing cation exchange product, without further purification, into a final container for therapeutic use,

wherein the albumin-containing cation exchange product is subjected to at least one step selected from the group consisting of buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning, before being placed into the said final container.

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Claim 80 (new): A process according to Claim 79 wherein the initial albumin solution contains glycosylated albumin and the glycosylated albumin is bound during the said cation exchange step.

Claim 81 (new): A process according to Claim 79 wherein prior to the cation exchange step the albumin solution undergoes at least one step selected from the group consisting of: (i) pH-adjustment; (ii) concentration; (iii) diafiltration; and (iv) conditioning by addition of a fatty acid.

Claim 82 (new): A process for purifying an albumin solution, the process comprising the steps of:

- (i) subjecting an albumin solution to a cation exchange chromatography step run in positive mode with respect to the albumin;
- (ii) collecting an albumin-containing cation exchange eluate;
- (iii) subjecting the cation exchange eluate to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (iv) collecting an albumin-containing anion exchange eluate;
- (v) subjecting the anion exchange eluate to an affinity chromatography step run in positive mode with respect to the albumin;
- (vi) collecting an albumin-containing affinity chromatography eluate;
- (vii) subjecting the affinity chromatography eluate to an affinity chromatography step run in negative mode with respect to the albumin and in positive mode with respect to glycoconjugates;
- (viii) collecting the albumin-containing affinity chromatography flow through;
- (ix) subjecting the affinity chromatography flow through to a cation exchange chromatography step run in negative mode with respect to the albumin;
- (x) collecting the albumin-containing cation exchange flow through;
- (xi) subjecting the cation exchange flow through to an anion exchange chromatography step run in negative mode; and
- (xii) collecting the albumin-containing anion exchange flow through from step (xi).

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Claim 83 (new): A process for purifying an albumin solution, the process comprising the steps of:

- (i). subjecting an albumin solution to a cation exchange chromatography step run in positive mode with respect to the albumin;
- (ii). collecting an albumin-containing cation exchange eluate;
- (iii). subjecting the cation exchange eluate to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (iv). collecting an albumin-containing anion exchange eluate;
- (v). subjecting the anion exchange eluate to an affinity chromatography step run in positive mode with respect to the albumin;
- (vi). collecting an albumin-containing affinity chromatography eluate;
- (vii). subjecting the affinity chromatography eluate to an affinity chromatography step run in negative mode with respect to the albumin and in positive mode with respect to glycoconjugates;
- (viii). collecting the albumin-containing affinity chromatography flow through;
- (ix). subjecting the affinity chromatography flow through to a cation exchange chromatography step run in negative mode with respect to the albumin;
- (x). collecting the albumin-containing cation exchange flow through;
- (xi). subjecting the cation exchange flow through to an anion exchange chromatography step run in positive mode; and
- (xii). eluting from the anion exchange matrix an anion exchange eluate.

Claim 84 (new): A process according to Claim 82 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

Claim 85 (new): A process according to Claim 83 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 83 (new): A process for purifying an albumin solution, the process comprising the steps of:

- (i). subjecting an albumin solution to a cation exchange chromatography step run in positive mode with respect to the albumin;
- (ii). collecting an albumin-containing cation exchange eluate;
- (iii). subjecting the cation exchange eluate to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (iv). collecting an albumin-containing anion exchange eluate;
- (v). subjecting the anion exchange eluate to an affinity chromatography step run in positive mode with respect to the albumin;
- (vi). collecting an albumin-containing affinity chromatography eluate;
- (vii). subjecting the affinity chromatography eluate to an affinity chromatography step run in negative mode with respect to the albumin and in positive mode with respect to glycoconjugates;
- (viii). collecting the albumin-containing affinity chromatography flow through;
- (ix). subjecting the affinity chromatography flow through to a cation exchange chromatography step run in negative mode with respect to the albumin;
- (x). collecting the albumin-containing cation exchange flow through;
- (xi). subjecting the cation exchange flow through to an anion exchange chromatography step run in positive mode; and
- (xii). eluting from the anion exchange matrix an anion exchange eluate.

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Claim 84 (new): A process according to Claim 82 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 85 (new): A process according to Claim 83 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 86 (new): A process for purifying an albumin solution, the process comprising the steps of:

- (i) subjecting an albumin solution to a cation exchange chromatography step run in positive mode with respect to the albumin;
- (ii) collecting an albumin-containing cation exchange eluate;
- (iii) subjecting the cation exchange eluate to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (iv) collecting an albumin-containing anion exchange eluate;
- (v) subjecting the anion exchange eluate to an affinity chromatography step run in positive mode with respect to the albumin;
- (vi) collecting an albumin-containing affinity chromatography eluate;
- (vii) subjecting the affinity chromatography eluate to an affinity chromatography step run in negative mode with respect to the albumin and in positive mode with respect to glycoconjugates;
- (viii) collecting the albumin-containing affinity chromatography flow through;
- (ix) subjecting the affinity matrix flow through to an anion exchange chromatography step run in negative mode with respect to the albumin;
- (x) collecting the albumin-containing anion exchange flow through from step (ix);
- (xi) subjecting the albumin solution purified by the anion exchange chromatography step to a cation exchange chromatography step run in negative mode with respect to the albumin; and
- (xii) collecting the albumin-containing cation exchange flow through.

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Claim 87 (new): A process for purifying an albumin solution, the process comprising the steps of:

- (i). subjecting an albumin solution to a cation exchange chromatography step run in positive mode with respect to the albumin;
- (ii). collecting an albumin-containing cation exchange eluate;
- (iii). subjecting the cation exchange eluate to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (iv). collecting an albumin-containing anion exchange eluate;

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- (v). subjecting the anion exchange eluate to an affinity chromatography step run in positive mode with respect to the albumin;
- (vi). collecting an albumin-containing affinity chromatography eluate;
- (vii). subjecting the affinity chromatography eluate to an affinity chromatography step run in negative mode with respect to the albumin and in positive mode with respect to glycoconjugates;
- (viii). collecting the albumin-containing affinity chromatography flow through;
- (ix). subjecting the affinity matrix flow through to an anion exchange chromatography step run in positive mode with respect to the albumin;
- (x). eluting from the anion exchange matrix an anion exchange eluate;
- (xi). subjecting the albumin solution purified by the anion exchange chromatography step to a cation exchange chromatography step run in negative mode with respect to the albumin; and
- (xii). collecting the albumin-containing cation exchange flow through.

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Claim 88 (new): A process according to Claim 86 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; addition of reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 89 (new): A process according to Claim 87 wherein any of the said purification steps are optionally preceded or followed by at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; addition of reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 90 (new): A process for purifying an albumin solution, the process comprising

- (1) subjecting the albumin solution to anion exchange chromatography in order to yield an albumin-containing anion exchange product;
- (2) subjecting the albumin-containing anion exchange product, with or without intervening purification steps, to cation exchange chromatography run in the negative mode with respect to the albumin to yield an albumin-containing cation exchange product; and

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(3) placing the albumin-containing cation exchange product, without further purification, into a final container for therapeutic use.

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Claim 91 (new): A process according to Claim 90 wherein the initial albumin solution contains glycosylated albumin and the glycosylated albumin is bound during the said cation exchange step

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Claim 92 (new): A process according to Claim 90 wherein the cation exchange step utilises a matrix which comprises immobilised sulfopropyl substituents as cation exchangers.

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Claim 93 (new): A process according to Claim 90 wherein the albumin solution that undergoes cation exchange chromatography has a pH of 4.5-6.0

Claim 94 (new): A process according to Claim 90 wherein the albumin solution that undergoes cation exchange chromatography has an albumin concentration of 10-250g.L⁻¹.

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Claim 95 (new): A process according to Claim 90 wherein the albumin solution that undergoes cation exchange chromatography has an octanoate ion concentration of 2-15mM.

59
Claim 96 (new): A process according to Claim 90 wherein prior to the cation exchange step the albumin solution undergoes at least one step selected from the group consisting of: (i) pH adjustment; (ii) concentration; (iii) diafiltration; and (iv) conditioning by addition of a fatty acid.

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Claim 97 (new): A process according to Claim 96 wherein prior to the cation exchange step the albumin solution undergoes conditioning by addition of an octanoate salt.

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Claim 98 (new): A process according to Claim 90 wherein the anion exchange step utilises a matrix which comprises immobilised dialkylaminoalkyl substituents as anion exchangers.

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Claim 99 (new): A process according to Claim 90 wherein the anion exchange step is run in the negative mode with respect to the albumin.

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Claim 100 (new): A process according to Claim 99 wherein the albumin solution which undergoes anion exchange chromatography has a pH of 4.0-5.2.

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Claim 101 (new): A process according to Claim 99 wherein the albumin solution which undergoes anion exchange chromatography has a conductivity of less than 4.0mS.cm^{-1} .

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Claim 102 (new): A process according to Claim 90 wherein the anion exchange step is run in positive mode with respect to the albumin

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Claim 103 (new): A process according to Claim 102 wherein the albumin solution which undergoes positive mode anion exchange chromatography has a pH of 6.0-8.0.

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Claim 104 (new): A process according to Claim 102 wherein the concentration of the albumin in the albumin solution which undergoes positive mode anion exchange chromatography is $10\text{-}100\text{g.L}^{-1}$

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Claim 105 (new): A process according to Claim 102 wherein the albumin solution which undergoes positive mode anion exchange chromatography has a conductivity of $1.0\text{-}1.5\text{mS.cm}^{-1}$.

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Claim 106 (new): A process according to Claim 102 wherein the albumin is eluted in the anion exchange step using a buffer comprising a compound having a specific affinity for albumin.

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Claim 107 (new): A process according to Claim 106 wherein the buffer comprises 20-90mM phosphoric acid salt.

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Claim 108 (new): A process according to Claim 102 wherein the albumin is eluted in the anion exchange step with a buffer of pH6.0-8.0.

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Claim 109 (new): A process according to Claim 90 wherein, prior to the cation exchange step, the albumin solution undergoes at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 110 (new): A process according to Claim 90 wherein the process is preceded by at least one step selected from the group consisting of: fermentation; primary separation; concentration; conditioning; cation exchange chromatography; anion exchange; and affinity chromatography.

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Claim 111 (new): A process for purifying an albumin solution, the process comprising

- (1) subjecting the albumin solution to anion exchange chromatography in order to yield an albumin-containing anion exchange product;
- (2) subjecting the albumin-containing anion exchange product, with or without intervening purification steps, to cation exchange chromatography run in the negative mode with respect to the albumin to yield an albumin-containing cation exchange product; and
- (3) placing the albumin-containing cation exchange product, without further purification, into a final container for therapeutic use,

wherein, prior to the cation exchange step, the albumin solution undergoes at least one step selected from the group consisting of: buffer exchange; concentration; dilution; dialysis; diafiltration; pH-adjustment; treatment with a reducing agent; decolouration treatment; heating; cooling; and conditioning.

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Claim 112 (new): A process according to Claim 111 wherein the initial albumin solution contains glycosylated albumin and the glycosylated albumin is bound during the said cation exchange step.

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Claim 113 (new): A process according to Claim 111 wherein prior to the cation exchange step the albumin solution undergoes at least one step selected from the group consisting of: (i) pH-adjustment; (ii) concentration; (iii) diafiltration; and (iv) conditioning by addition of a fatty acid.